

Anti-COPE Antibody Picoband™ (monoclonal, 9B6)

Catalog # ABO14882

Product Information

Application	WB, IHC, IF, ICC, FC
Primary Accession	O14579
Host	Mouse
Isotype	Mouse IgG2a
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-COPE Antibody Picoband™ (monoclonal, 9B6) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500 µg/ml.

Additional Information

Gene ID	11316
Other Names	Coatomer subunit epsilon, Epsilon-coat protein, Epsilon-COP, COPE
Calculated MW	34482
Application Details	Western blot, 0.1-0.5 µg/ml Immunohistochemistry (Paraffin-embedded Section), 0.5-1 µg/ml Immunocytochemistry/Immunofluorescence, 5 µg/ml Flow Cytometry, 1-3 µg/1x10 ⁶ cells
Subcellular Localization	Golgi apparatus membrane. Peripheral membrane protein. Cytoplasmic side. Cytoplasm. COPI-coated vesicle membrane.
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
Clone Names	Clone: 9B6
Immunogen	E. coli-derived human COPE recombinant protein (Position: E80-A308). Human COPE shares 89.5% amino acid (aa) sequence identity with mouse COPE.
Cross Reactivity	No cross-reactivity with other proteins.
Storage	Store at -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freeze-thaw cycles.

Protein Information

Name COPE

Function The coatomer is a cytosolic protein complex that binds to dilysine motifs and reversibly associates with Golgi non-clathrin-coated vesicles, which further mediate biosynthetic protein transport from the ER, via the Golgi up to the trans Golgi network. The coatomer complex is required for budding from Golgi membranes, and is essential for the retrograde Golgi-to-ER transport of dilysine-tagged proteins. In mammals, the coatomer can only be recruited by membranes associated with ADP-ribosylation factors (ARFs), which are small GTP-binding proteins; the complex also influences the Golgi structural integrity, as well as the processing, activity, and endocytic recycling of LDL receptors (By similarity).

Cellular Location Cytoplasm. Golgi apparatus membrane; Peripheral membrane protein; Cytoplasmic side. Cytoplasmic vesicle, COPI-coated vesicle membrane; Peripheral membrane protein; Cytoplasmic side. Note=The coatomer is cytoplasmic or polymerized on the cytoplasmic side of the Golgi, as well as on the vesicles/buds originating from it.

Background

Coatomer subunit epsilon is a protein that in humans is encoded by the COPE gene. The product of this gene is an epsilon subunit of coatomer protein complex. Coatomer is a cytosolic protein complex that binds to dilysine motifs and reversibly associates with Golgi non-clathrin-coated vesicles. It is required for budding from Golgi membranes, and is essential for the retrograde Golgi-to-ER transport of dilysine-tagged proteins. Coatomer complex consists of at least the alpha, beta, beta', gamma, delta, epsilon and zeta subunits. Alternatively spliced transcript variants encoding different isoforms have been identified.

Images

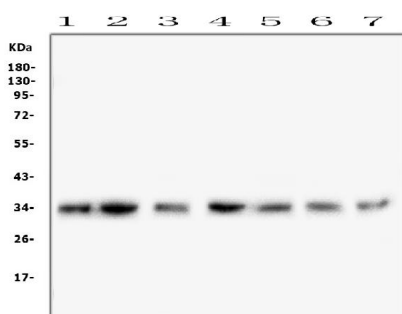


Figure 1. Western blot analysis of COPE using anti-COPE antibody (M04544).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.

Lane 1: human HEK293 whole cell lysates

Lane 2: human HepG2 whole cell lysates

Lane 3: human placenta tissue lysates

Lane 4: human Caco-2 whole cell lysates

Lane 5: human A549 whole cell lysates

Lane 6: human PANC-1 whole cell lysates

Lane 7: human SW579 whole cell lysates

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-COPE antigen affinity purified monoclonal antibody (Catalog # M04544) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for COPE at

approximately 34KD. The expected band size for COPE is at 34KD.

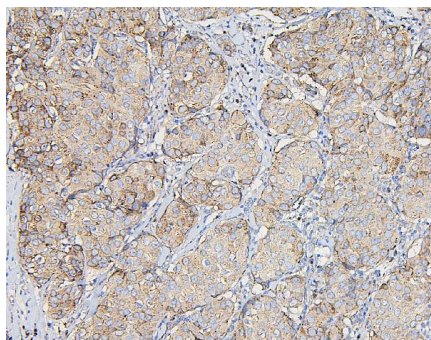


Figure 2. IHC analysis of COPE using anti-COPE antibody (M04544). COPE was detected in paraffin-embedded section of human mammary cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml mouse anti-COPE Antibody (M04544) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

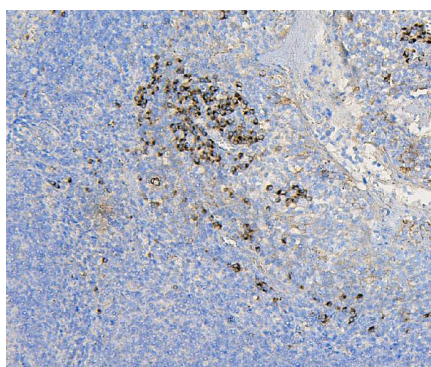


Figure 3. IHC analysis of COPE using anti-COPE antibody (M04544). COPE was detected in paraffin-embedded section of human tonsil tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml mouse anti-COPE Antibody (M04544) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

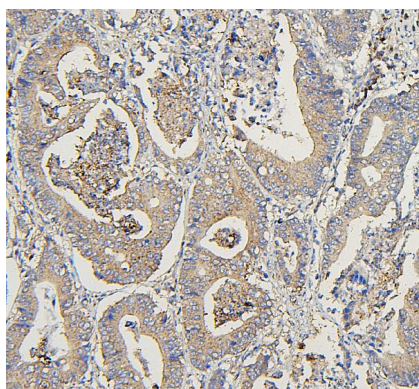


Figure 4. IHC analysis of COPE using anti-COPE antibody (M04544). COPE was detected in paraffin-embedded section of human intestinal cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml mouse anti-COPE Antibody (M04544) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

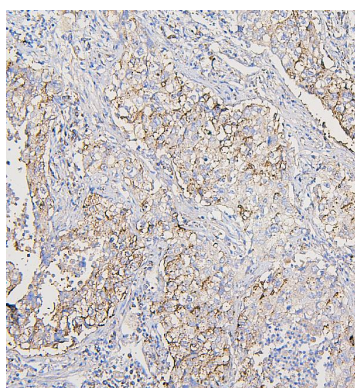


Figure 5. IHC analysis of COPE using anti-COPE antibody (M04544). COPE was detected in paraffin-embedded section of human lung cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml mouse anti-COPE Antibody (M04544) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

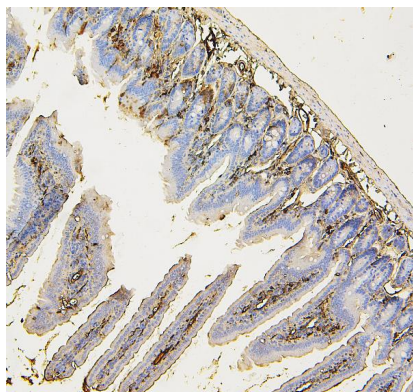


Figure 6. IHC analysis of COPE using anti-COPE antibody (M04544).

COPE was detected in paraffin-embedded section of mouse intestine tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-COPE Antibody (M04544) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

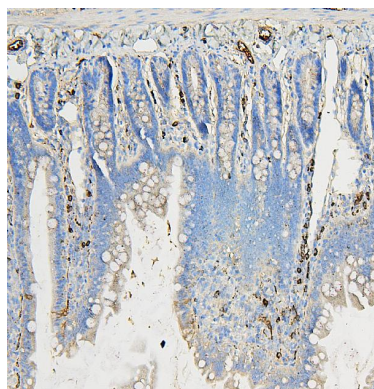


Figure 7. IHC analysis of COPE using anti-COPE antibody (M04544).

COPE was detected in paraffin-embedded section of rat intestine tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-COPE Antibody (M04544) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

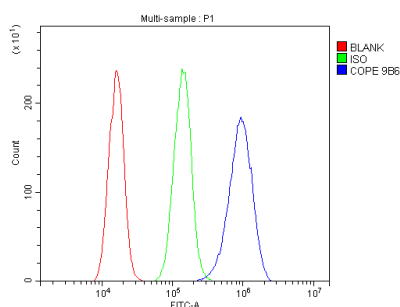


Figure 8. Flow Cytometry analysis of A431 cells using anti-COPE antibody (M04544).

Overlay histogram showing A431 cells stained with M04544 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-COPE Antibody (M04544, 1 µg/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

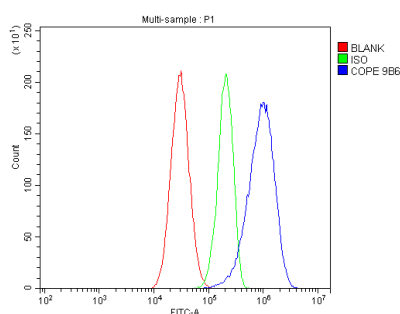
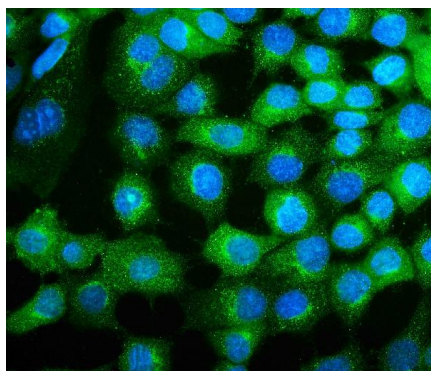


Figure 9. Flow Cytometry analysis of HepG2 cells using anti-COPE antibody (M04544).

Overlay histogram showing HepG2 cells stained with M04544 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-COPE Antibody (M04544, 1 µg/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Figure 10. IF analysis of COPE using anti-COPE antibody (M04544).

COPE was detected in immunocytochemical section of A431 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15



mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-COPE Antibody (M04544) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

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